Antibody Responses to *Aspergillus fumigatus* Allergens in Patients with Cystic Fibrosis

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Key Words

Aspergillus fumigatus
Cystic fibrosis
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Introduction

Fungi of the genus *Aspergillus* are associated with a spectrum of human diseases including asthma, allergic bronchopulmonary aspergillosis (ABPA), aspergilloma and cystic fibrosis (CF). We have previously reported that CF patients have a significantly higher prevalence of IgE antibody (Ab) to *Aspergillus fumigatus* (60%) than patients with asthma (6%). In addition, most CF patients (84%) had serum IgG Ab to *A. fumigatus* allergen Asp f 1. A subset of CF patients fulfilled the immunologic criteria for ABPA, including some children < 5 years old [1]. Asp f 1, which is a major [\8-kO](#) *A. fumigatus* allergen, is a member of the mito-gillin family of cytotoxins [2]. The complete nucleotide sequence of Asp f 1 has been determined [3], and recombinant Asp f 1 with IgE-binding activity has been produced [4]. In addition, Asp f 1 causes proliferative T cell responses in patients with ABPA [5]. Our studies suggested that CF patients are frequently colonized with *A. fumigatus*, and that this colonization may contribute to the lung damage in some CF patients.

Methods and Results

We have extended the analysis of *A. fumigatus* IgG and IgE Ab responses to a group of 119 CF patients (age 10 months[^]-6 years). IgG Ab was measured by antigen-binding RIA using ¹²⁵I-labeled Asp f 1, and IgE Ab to *A. fumigatus* was quantitated by radioallergosorbent test. Eighty-nine percent of the patients had detectable IgG to Asp f 1, and 14% fulfilled the immunologic criteria for ABPA. The RIA using ¹²⁵I-labeled Asp f 1 was also used to monitor IgG Ab levels following antifungal therapy. Treatment of two CF patients with itraconazole over periods of 7 and 12 months resulted in a significant decrease in IgG Ab levels. In addition, a 6-year follow-up of a patient who presented with invasive aspergilloma and who was treated with surgery and amphotericin B [6] has shown a steady decrease (up to 65%) in the levels of IgG Ab to Asp f 1.

To further identify *A.fumigatus* allergens, a cDNA library was prepared from *A.fumigatus* mycelial mRNA. IgE Ab of a serum pool obtained from 8 patients with CF or ABPA was used to screen the library. Three positive clones were isolated, containing inserts of 0.8-1.3 kb, and sequencing of the clones is in progress. Results of plaque immunoassays showed that 40-70% of CF patients had IgE Ab to these allergens.

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Discussion

Analysis of a large panel of serum from CF patients showed that a high percentage have IgG Ab to Asp f 1, a major *A. fumigatus* allergen. In keeping with previous studies, we found a prevalence of 14% of patients with immunologic criteria for ABPA. The results suggested that IgG Ab to Asp f 1 is a good marker for *A. fumigatus* colonization and can be used for evaluating antifungal treatment. Previous in vitro studies have shown that Asp f 1 is secreted into the culture medium upon germination of *A. fumigatus* spores, and that undetectable or very little Asp f 1 is present in spores or hyphae [3, 7]. It is possible that Asp f 1 may contribute to lung damage through IgE-mediated inflammation and/or direct cytotoxicity to lung tissue. Our results suggest that *A. fumigatus* in addition to *Pseudomonas aeruginosa* and *Staphylococcus aureus* should be considered as causes of lung disease in some patients with CF.

Although Asp f 1 appears to be a good marker for *A. fumigatus* colonization, there is evidence that *Aspergillus* spp. produce other allergens. Using IgE Ab from CF serum, we have isolated three cDNA clones from an *A. fumigatus* library. Identification and sequencing of *A. fumigatus* allergens other than Asp f 1 will allow further studies on the pathogenesis of *Aspergillus*-related diseases.

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